consecutive hybrid quadruplexes. With addition of crowding mimetics (Ficoll 70; Ficoll 400; and PEG 200 (20 and 40% w/v)) to the HT4 sequence, CD studies show a conformational switch to an antiparallel structure in Ficoll 400 (40% w/v) and a parallel conformation in PEG 200 (40% w/v), with no conformational changes observed in Ficoll 70. For HT8, a similar effect was observed, but in addition an anti-parallel conformation was obtained in Ficoll 70 (40% w/v), which suggests an enhanced sensitivity to dehydration for the HT8 sequence. Additionally, thermal CD melting studies showed stabilization of HT8 conformation in Ficoll compared with HT4. With binding of NMM to HT4 in 20% (w/v) crowding mimetics a shift to the parallel conformation was observed. However, the antiparallel (more dehydrated) conformation for HT8 is promoted in Ficoll 400. We plan more detailed environmental studies of NMM binding to qDNA under crowding conditions using fluorescence spectroscopy.
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2171-Pos Board B190
Zipper Like Structures Possible Intermediates to Assemble Duplex Mediated G-Quadruplex DNA
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In this paper we report the detailed characterization at the atomic level by molecular mechanics of Guanine-rich DNA that forms zipper-like structures. These duplexes in vitro have shown to associate into G-quadruplex that are conformed by two duplexes. We want to known at the atomic level how the association process works, but the structure of the duplex for these sequences has not been established. This is a preliminary work to be able to model the formation of the zipper structure (G-quadruplex mediated structure). Such process has been hypothesized to work at sites rich in Gs and be involved in process like meiosis, pairing homologues chromosomes and the structural function of telomeres. Models were constructed and these structures minimized and equilibrated, allowing analysis via molecular dynamics to understand the factors that determines the most stable structure. Preliminary analysis studying the intrinsic chemical stability showed that the presence of positively charged ions near the Gs-rich region of the studied sequences is critical to the stability of these DNA structures. It has been shown that simulating divalent cations can be challenging and that the force fields have to be tested for non canonical DNA conformations. This particular structure has a normal close to B-DNA section but the ''zipper-like'' is definitively a region that needs to be carefully studied to check the adequacy of the modeling tools used. The dynamics of these molecules show us that zipper structures have more stability with sodium ions in simulated conditions. In summary, the results allow a better understanding of this system at the molecular level, allowing us to predict the likelihood of formation of these structures in vivo as well the features of the sequences that could fold as zipper-like structures.

2172-Pos Board B191
Existence and Possible Function of Buckled DNA in Tailed DSDNA Bacteriophage Portals
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Tailed double-stranded DNA (dsDNA) bacteriophages control genome packaging and ejection from their viral capsids using a portal system. The portal vertex serves as a docking site for the viral ATPase packing motor which translocates DNA through the central channel during packaging. In the case of several well-studied bacteriophages including T7 and ε15, the central channel extends into the capsids interior by a core assembly of stacked, cylindrical protein subunits. Often possessing different internal diameters, these protein subunits can create large cavities in an otherwise straight channel. The height of these cavities are typically within 10-20% of the DNA persistence length and, in the case of T7, the cavity is 50Å tall and 110Å wide. Given that these cavities exist upstream of the packing motor, we postulate that they allow DNA to buckle under large packing forces (~100 pN). A cryo-EM reconstruction of φ29 revealed that DNA buckles in a toroidal supercoil within a cavity only 3.5 times wider and 2.5 times taller than the width of DNA. A recent reconstruction of bacteriophage P22 also revealed DNA density inside its portal cavity that is over twice the width of dsDNA, suggesting that DNA may be compressed into a highly-bent supercoil. using analytic and numerical approaches, we compute the forces required for and during DNA buckling in bacteriophages T7 and P22. We demonstrate that DNA can indeed buckle and that the buckled conformation subsequently pushes outward on the cavity. Thus, the buckle could mechanically initiate a conformational change in the portal protein to provide the head-full signal.

2173-Pos Board B192
Coarse-Grained Model DNA with Explicit H-Bonding and Implicit Solvent
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We present a coarse-grained model for DNA that is intended to function realistically at the level of individual bases. The model is composed of residues with up to eight coarse-grained beads each, which is sufficient for DNA-like base stacking and base-base recognition by hydrogen bonding. The beads interact by means of short-ranged pair potentials and a simple implicit solvent model. Movement is simulated by Brownian dynamics without hydrodynamic coupling. The main stabilizing forces are base stacking and hydrogen bonding, as modified by the effects of solvation. Complementary double-stranded chains of such residues form stable double helices over long runs (~10 μs) at or near room temperature, with structural parameters close to those of B-form DNA. Most mismatched chains or mismatched regions within a complementary molecule melt and become disordered. Long-range fluctuations and elastic properties, as measured by bending and twisting persistence lengths, are close to experimental values. Single-stranded chains are flexible, with transient stretches of free bases in equilibrium with globules stabilized by intrastrand stacking and hydrogen bonding. Model DNAs in covalently closed loops form right or left-handed supercoils, depending on the sign of overtwist or undertwist. Short stem-loop structures melt at elevated temperatures and reanneal when the temperature is carefully lowered. Overall, most qualitative properties of real DNA arise naturally in the model from local interactions at the base pair level.

2174-Pos Board B193
Molecular Dynamics Study of Cobalt(III) Hexammine Counter-Ion Distributions around B-DNA and A-RNA Duplexes
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Multivalent ions are known to induce strong attractive forces between DNA strands resulting in DNA condensation. At the same time, short double-stranded (ds) RNA helices resist condensation by trivalent Cobalt hexammine (Cohex) in the DNA condensing ionic conditions. To explore the factors that could lead to this difference in condensation, we have carried out a set of explicit solvent molecular dynamics simulations of 25 base pairs canonical B-DNA and A-RNA duplexes under different salt conditions. Several mixtures of monovalent (Na, Cl) and trivalent (Cohex) ions are considered. The results of simulation show that Co-hex ions effectively displace monovalent Na ions from the major groove of dsRNA binding to the RNA phosphate oxygens. The ions are buried within the major groove with a distribution peak at 7.3 Angstrom from the helix axis. their density rapidly decreases and becomes negligible at 11 A from the axis. In contrast to that, the distribution of Cohex around B-DNA is shifted outside the helix with a peak at 13.3 A from the helix axis. The ions prefer to bind to the phosphate groups on the outer surface of B-DNA. The observed difference in Cohex distributions around B-DNA and A-RNA is in agreement with the proposed explanation of the resistance of RNA to condensation due to a difference in Cohex binding to DNA and RNA (L.Li et al., PRL 106, 108101 (2011)).

2175-Pos Board B194
Dynamic NMR Studies Provide Insight into Sequence Dependent Binding Affinities
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The ability of molecules to bind DNA plays an important role in many biological processes including, transcription, regulation, replication, repair. Understanding the factors that influence binding affinity is therefore essential to our understanding of these processes.

In this study the effect of sequence context on the binding affinity and conformation of DNA dodecamers are explored. The Cre binding site (ACGT) was studied with a number of different flanking sequences. 31P-NMR was used to determine the conformational state (BI/BII ratio) of each step in the phosphate backbone. Importantly, the conformation of the binding site, the center tetrad which was not changed, varied significantly based on the sequence context.
Fluorescence titration with the DNA intercalator 7-Aminoactinomycin D was performed to provide biological context to our conformation results. We found that the backbone conformation alone cannot explain the binding affinities in the dodecamers studied. Thus, we turned our attention to base pair dynamics. Preliminary results show that differences in binding affinities in sequences with similar backbone conformations can be explained by differential base pair opening rates.

2176-Pos Board B195
Differential Binding Response of Copper(II)-NSAIDs to Variation in Base Sequences in the DNA Backbone
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Drugs belonging to the Non-Steroidal Anti-Inflammatory (NSAID) group are not only used as anti-inflammatory, analgesic and anti-pyretic agents, but also show anti-cancer effects. Complexing it with a bioactive metal like copper show an enhancement in their anti-cancer effects, whose exact mechanism of action is not yet fully understood. As a possible molecular mechanism behind the anticancer effects of Cu(II)-NSAIDs, our group has shown for the first time, that copper complexes of two NSAIDs belonging to oxicam chemical group namely meloxicam and piroxicam can directly bind to the DNA backbone. Elucidating base sequence specific interaction of Cu(II)-NSAIDs to the DNA will provide information on their possible binding sites in the genome sequence. We have identified the differences in the interaction of Cu(II)-piroxicam and Cu(II)-meloxicam with hompolymeric polydA-polydT and alternating polydA-dT as well as with homopolymeric polydG-polydC and alternating polydG-dC. Differential response of the two Cu(II)-NSAIDs to the subtle variation in backbone structures of polydA-polydT and polydA-dT, was expressed in the difference in their binding parameters. Both the complexes show similar binding modes with polydA-dT which is intercalative, but for polydA-polydT, the results point to a mixed mode of binding. Even for the mixed mode, there is a significant difference in the binding of Cu(II)-piroxicam and Cu(II)-meloxicam to polydA-polydT. However, for the GC rich sequences, Cu(II)-NSAIDs show strong binding affinity to both polydG-dC and polydG-polydC. The role reversal of Cu(II)-meloxicam from a strong binder of polydG-dC to a weak binder of polydG-polydC, while Cu(II)-piroxicam changes from a strong binder of polydG-polydC to a weak one of polydG-dC, point to the sensitivity of these complexes to changes in the backbone structures/hydration.
Hence, Cu(II)-NSAID complexes are highly sensitive to differences in backbone sequences.

2177-Pos Board B196
DNA and RNA Charge Transport Effect of Sequence, Stacking, Structure and Hg Incorporation
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In this work experimental and theoretical approaches have been combined for unique study and comparison of charge transfer efficiency in standard and modified (Hg incorporated) DNA duplexes and DNA/RNA hybrids. Specific mechanism proposed to account for the on-chain oligonucleotide charge transport is based on polaron formation-charge is transported coherently by long-distance tunneling (superexchange). A key parameter in this approach is how the bridge orbitals of the intervening DNA bases couple the charge donor and acceptor - this is strongly controlled by specific oligomeres structures and conformation. The RNA/DNA system flexibility also improves the conditions for polaron creation and thus may facilitate rapid (coherent) charge transport in the DNA/RNA hybrids. All our observations demonstrate that charge transfer characteristics of oligonucleotide duplexes are able to reveal the presence of structure changes in the sequence, which may help to elucidate possible disturbance identification in the cell or even in potential technological applications.

2178-Pos Board B197
Determining the Conformations of Porphyrin Dimers and Dinucleotide-Substituted DNA Constructs by 2-Dimensional Fluorescence Spectroscopy
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Exciton-coupled dimers exhibit spectroscopic properties that are very sensitive to dimer conformation, making them useful probes of local structure when incorporated into systems of biological interest. Numerous spectroscopic techniques can be used to study such dimers, including linear absorption spectroscopy and circular dichroism. 2-Dimensional Fluorescence Spectroscopy (2D FS) is a technique that involves exciting a sample with a series of four ultrafast laser pulses and measuring fluorescence as a function of the time delays between pulses, yielding spectra that sensitively reflect the conformations of exciton-coupled dimers. I will describe studies in which 2D FS was combined with linear absorption spectroscopy to ''solve the structure'' of a dimer of zinc tetraphenylporphyrin embedded in a phospholipid bilayer membrane. It was found that the dimer adopts a mixture of folded and extended conformations in the membrane and that higher temperatures favor the folded conformation. A thermodynamic analysis revealed that this temperature-dependent conformational shift is driven by the increase in entropy of the lipid side chains upon folding of the porphyrin dimer, analogous to the hydrophobic effect that drives the folding of many proteins. I will also describe experiments in which 2D FS was used to study nucleic acid constructs substituted with two adjacent fluorescent base analogues. In a similar manner to the porphyrins, the exciton coupling between base analogues allows 2D FS signals to be combined with other spectroscopic signals to determine the local conformation of the nucleic acid. Ongoing experiments aim to use this technique to study the local structure and ''real-time'' dynamics of the nucleic acid components of DNA-protein complexes in solution.

2179-Pos Board B198
Ionic Microenvironmental effects on Triplex DNA Stabilization
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DNA structure and conformation are influenced by metal ions, polyamines, and the microenvironment. Positively charged ions stabilize DNA and RNA structures, and provoke conformational transitions. In poly(purine).poly(pyrimidine) sequences, triplex DNA formation is facilitated by metal ions, polyamines and several other ligands. We studied the effects of mono, and di-valent metal ions, and ammonium salts on the stability of triple- and double-stranded structures formed from poly(dA) and poly(dT) by measuring their respective melting temperatures. In the presence of metal ions, the absorbance versus temperature profile showed two transitions: Tm1 for triplex to duplex and single stranded DNA, and Tm2 for duplex DNA melting to single stranded DNA. Monovalent cations (Li^+, Na^+, K^+, Rb^+, Cs^+ and $NH4^+$) promoted triplex DNA at concentrations $\geq$ 150 mM. The triplex DNA melting temperature varied from 49.8 °C in the presence of 150 mM Li^+ to 30.6 °C in the presence of 150 mM K^+. Among ammonium compounds, $NH4^+$ was the most effective ion in stabilizing triplex DNA and its efficacy decreased with increasing substitution of the hydrogen atoms with bulky alkyl groups. Divalent cations were 1000-fold more efficacious than monovalent ions in stabilizing triplex DNA. All positively charged cations increased the melting temperature of duplex DNA. using 1/Tm1/1/Tm2 versus ln [ion concentration] plots, we calculated the amount of cation release and appropriate thermodynamic parameters, on triple/duplex melting. Circular dichroism spectroscopic studies showed distinct conformational changes in triplex DNA stabilized by alkali metal and ammonium ions. The results suggest ion specific effects on triplex DNA stabilization. Our results might be useful in developing triplex forming oligonucleotide based gene silencing techniques.

2180-Pos Board B199
Comparing the effects of Different Osmolytes in the B-To-Z DNA Transition
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